

Last copepodid, female.—The outline of the body is similar to that of the male. The average measurements for both copepodid and adult females, corresponding to those given for the male, are indicated in Table 1. The pair of sensilla on each side of the last metasomal somite (Fig. 20) are smaller than the prominent ones of the adult (Fig. 21) which are highly characteristic of *D. sanguineus*. The urosome in both instars consists of three somites. The genital segment of the copepodid (Fig. 20) is simple, but that of the adult is elongate and asymmetrical (Fig. 21).

The right and left antennules are similar in both instars, resembling the nonprehensile appendages of the males. The species is of the "little setaceous" type, having only one seta on segments 11 and 13-19. The antennae and mandibles differ in the two instars in the same way as in the male. The first and second maxillae, maxillipeds, and first through fourth legs are similar to those of the male.

The fifth leg in the copepodid (Fig. 22) differs from the adult in the presence of the lateral seta of the second exopodite podomere. The prominent claw of the adult (Fig. 23) is weakly developed. In specimens about to molt the form of the adult female may be seen within (Fig. 24).

BOTANY.—*A study of the roots of Pinus virginiana in relation to certain Hymenomyces suspected of being mycorrhizal.* EDWARD HACSKAYLO, George Washington University. (Communicated by William W. Diehl.)

The occurrence of ectotrophic mycorrhizae on *Pinus virginiana* Mill., a common conifer of the eastern United States, has been referred to by Henry (1), Kelley (2), McComb (4), and McDougall (5), but no mention was made concerning the identity of the fungi involved in the relationship with this tree species. Inasmuch as fungal cultures could be obtained from sporophores collected from a stand of pine, it would be possible to determine experimentally the identity of some of those species associated in the mycorrhizae of *P. virginiana*. This could be accomplished by subjecting seedlings germinated under aseptic conditions to simple inoculation tests using pure cultures of each fungus suspected

The principal points of difference between the last copepodid and the adult stages, aside from changes in body size and proportions, are found in both sexes in the development of the sensilla of the last metasomal somite, in the number of terminal setae on the endopodites of the antennae and the mandibles, and in the form of the fifth legs. The male is further distinguished by the number of somites in the urosome, and by the structure of the right antennule. The female differs conspicuously in the development of the genital segment.

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because of its constant association with the pine stands.

Sporophores of several Hymenomyces were collected during the summer and fall of 1949 from two nearly pure stands of *Pinus virginiana* occurring in Virginia near Washington, D. C. The following fungi were identified: *Amanita verna* (Bull.) Quel., *Boletus americanus* Pk., *Boletus* sp., *Clavaria pulchra* Pk., *Lactarius chrysothorus* Fr., *L. piperatus* (Scop.) Fr., *Tricholoma equestre* (L.) Quel., and *T. portentosum* Fr. From young sporophores of the above, tissue fragment cultures were obtained on a 50-50 mixture of commercial potato dextrose and malt dextrose agars.

Seeds of *Pinus virginiana*, purchased from the Herbst Brothers Seed Co., of New York, were surface-sterilized by soaking them for five minutes in a 1:1000 aqueous solution of bichloride of mercury. Following this treatment, the seeds were washed four times with sterile deionized water and then placed on a 2% (Ben Venue¹ green plant) agar medium containing the following amounts of salts per liter: MgSO₄, 1.2 gm; (NH₄)₂SO₄, 0.2 gm; Ca(NO₃)₂, 0.8 gm; KH₂PO₄, 0.7 gm and a trace of CuSO₄. They were germinated aseptically in Petri dishes and urine specimen bottles in a room at approximately 24°C. not exposed to direct sunlight.

In January and February, 1950, 100 pot cultures of pine seedlings were inoculated with the various fungi in culture, and 16 uninoculated controls were prepared. The methods used were similar to those described by McArdle (3) modified as follows. Each 3-inch pot was equipped with a glass subirrigation tube in order to avoid washing surface contaminants into the substrate during watering and applying nutrient solution to the cultures. The end of tube exposed to the air was covered with a glass vial when not in use. The system tended to reduce the amount of moisture on the substrate surface and consequently was not favorable for air-borne contaminants. The nutrient solution used was the same as prescribed by McArdle (3).

The pot cultures were maintained in the greenhouse where growth was generally favorable. There was, however, some variation in the amount of shoot growth among the different cultures.

In April 1950, examination of some of the roots showed profuse dichotomy of the short roots, this having developed in less than four months in cultures containing mycelium of *Amanita verna*. Dichotomy was also present to a lesser extent in the cultures containing mycelium of *Lactarius piperatus* and *Tricholoma portentosum*, but not in the pots con-

taining the other fungi noted above. In free-hand sections of these dichotomous roots, it was found that initials of ectotrophic mycorrhizae had developed since mantles were present. Hyphal penetration was not evident as a well-developed Hartig net. Roots of eight of the control plants were examined and neither dichotomy nor fungus association was found.

The roots of seedlings that were maintained and examined after a period of six to eight months did not appear to be growing actively during late spring and summer. There were neither mycorrhizal nor nonmycorrhizal young roots and very few older structures that resembled mycorrhizae. High temperatures up to 46°C. noted in the greenhouse after April 1 may have inhibited mycorrhizal formation and root development.

These results indicate that *Amanita verna*, *Lactarius piperatus*, and *Tricholoma portentosum* are possibly involved in the mycorrhizae of *Pinus virginiana*. No such indications, however, were found with *Boletus americanus*, *Boletus* sp., *Clavaria pulchra*, *Lactarius chrysorrheus*, and *Tricholoma equestre* under the conditions of the experiment. None of the roots of the control plants were found to be associated with a fungus. This work has revealed some of the problems that must be solved in future experimentation and is a forerunner of research now under way.

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